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(54) Title: POTENTIATION OF PRODRUG EFFICACY

(57) Abstract: The invention provides methods for statistically significantly potentiating the activity of a prodrug without producing significant side effects, the method comprising co-administering a polyanion with the prodrug.

POTENTIATION OF PRODRUG EFFICACY

BACKGROUND OF THE INVENTION

Field of the invention

The invention relates to the therapeutic use of prodrugs. More particularly, the invention relates to methods for potentiating the efficacy of prodrugs.

Summary of the related art

Many potential pharmaceutical agents fail to be used therapeutically due to excessive toxicity or limited bioavailability. In some instances, these limiting factors can be ameliorated by modifying the pharmaceutical agent to create a prodrug. The prodrug is then converted by the body into the pharmaceutically active substance.

International application number PCT/US97/14751 discloses the manufacture of oligonucleotide prodrugs having ester or amide modifications that cover a non-bridging oxygen of the phosphodiester linkage. Kuhn, Oncology, Supplement No. 6, 39-42 (1998) discloses that CPT-11 (Camptosar) is an antineoplastic prodrug that is converted by carboxylesterase activity in the liver and other tissues to the active agent SN-38. Cerosimo, The Annals of Pharmacotherapy 32: 1324-1333 (1998) teaches that the parent compound of CPT-11, camptothecin, was unable to be developed as a pharmaceutical due to severe toxicity.

Due to the presence of carboxylesterases and amidases in the liver and other tissues, the ability to make prodrugs which have added ester or amide groups is a generalizable phenomenon. However, these compounds generally retain at least some of the toxicity of the parent compound, due to rapid hydrolysis of the prodrug. Kuhn, *supra*, discloses that SN-38, the active metabolite of CPT-11 still causes diarrhea, which is the limiting toxicity of the parent compound, camptothecin.

Thus, there is a need for methods to administer prodrugs in a manner that maximizes their efficacy while avoiding significant toxicity. Ideally, such methods

should affect the manner in which the body processes prodrugs, and thus would be applicable to a broad range of prodrugs.

BRIEF SUMMARY OF THE INVENTION

The invention provides methods for administering prodrugs in a manner that maximizes their efficacy, and thus allows lower, less toxic dosages to be used. The methods according to the invention act through a variety of mechanisms that modulate the ability of the body to process the prodrug to the active compound and its ability to clear either the prodrug or the active compound, and are thus applicable to a broad range of prodrugs.

The methods according to the invention comprise co-administering to the patient a prodrug, preferably an ester or amide prodrug, and a polyanion, preferably a polysulfate. Preferred prodrugs include, without limitation, esters or amides of anti-cancer drugs, such as Camptosar and Camtosar analogs. Preferred polyanions include, without limitation, heparin, dextran sulfates, suramin sulfates, cyclodextrin sulfates and oligonucleotides, especially oligonucleotide phosphorothioates or phosphorodithioates.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 shows survival times for mice in the HCT116 study using Oligo 1.

Figure 2 shows survival times for mice in the HCT116 study using Oligo 2.

Figure 3 shows a Kaplan-Meier survival plot for mice in the HCT116 study using Oligo 1.

Figur 4 shows a Kaplan-Meier survival plot for mice in the HCT116 study using Oligo 2.

Figure 5 shows the time of administration of oligonucleotide effect on survival times for mice.

Figure 6 shows the efficacy of oral administration of oligonucleotides reflected in survival times for mice.

DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENTS

The invention relates to the therapeutic use of prodrugs. More particularly, the invention relates to methods for potentiating the efficacy of prodrugs. The patents and publications recited herein reflect the level of knowledge in this field and are hereby incorporated by reference. Any conflict between the teachings of these references and the present specification shall be resolved in favor of the latter.

The invention provides methods for administering prodrugs in a manner that maximizes their efficacy, and thus allows lower, less toxic dosages to be used. The methods according to the invention act through a variety of mechanisms that modulate the ability of the body to process the prodrug to the active compound and its ability to clear either the prodrug or the active compound, and are thus applicable to a broad range of prodrugs.

In a first aspect, the invention provides methods for statistically significantly potentiating the activity of a prodrug without producing significant side effects, the method comprising co-administering a polyanion with the prodrug. As used herein, a "prodrug" is a compound comprising an active compound covalently linked to another moiety by a cleavable linkage, wherein the pharmacological activity of the active compound is greater than the pharmacological activity of the prodrug, and wherein the active compound is produced in the body by cleavage of the cleavable linkage. An "active compound" is a molecule having a pharmacological activity. A "pharmacological activity" is an activity that is useful in the treatment of one or more disease or disease symptom. A "moiety" is a chemical group or structure. A "cleavable linkage" is a covalent bond that can be cleaved by an enzymatic activity in the body. The term "without producing significant side effects" means that any signs or

symptoms of toxicity that are observed in the presence of the polyanion are not greater than those observed in the absence of the polyanion to an extent that would preclude the combination of the prodrug and the polyanion from obtaining regulatory approval. The term "co-administration" is intended to include treatment regimens in which either the prodrug or the polyanion is continued after the cessation of the other agent.

Preferred prodrugs include amides and esters of active compounds. Such active compounds include, without limitation, anticancer chemotherapeutics, antinflammatory agents, antiinfectious agents, antiviral agents and cardiovascular drugs. Numerous prodrugs are well known in the art (see, *e.g.*, Singh *et al.*, *J. Sci. Ind. Res.* 55: 497-510 (1996)). A non-limiting example of preferred active compounds is SN-38. Specific non-limiting examples of preferred prodrugs include Camptosar ((7-ethyl-10-(-4-piperidinol)-1-piperidnocarbonyloxy-camptothecin; CPT-11) and Captosar analogs and foscarnate. The moiety that is cleaved from the prodrug may preferably be selected from esters and alpha-acyloxyalkyl esters (for carboxy functionalities); amides, esters, carbonate sters, phosphate esters, ethers and alpha-acyloxyalkyl ethers (for hydroxy functionalities); thioesters, alpha-acyloxyalkyl thioiesters and disulfides (for sulfhydryl functionalities); ketals, imines, enol esters, oxazoladines, and thiazolidines (for carbonyl functionalities); amides, carbamates, imines enamines N-Mannich bases, and N-acyloxyalkoxycarbonyl derivatives (for amino functionalities); N-acyloxyalkyl derivatives (for quarternary amino functionalities); N-sulphonyl imidates (for ester or sulfonamido functionalities); N-Mannich bases (for NH-acidic functionalities); and N-acyloxyalkyl derivatives (for heterocyclic amino functionalities).

Preferred polyanions include, without limitation, polysulfates and oligonucleotides. Preferred polysulfates include heparin, dextran sulfates, suramin sulfates, cyclodextrin sulfates and oligonucleotide phosphorothioates or phosphorodithioates. For purposes of the invention, the term "oligonucleotide"

includes polymers of two or more deoxyribonucleotide, or any modified nucleoside, including 2′-halo-nucleosides, 2′-O-substituted ribonucleosides, 3′-O-substituted nucleosides, deazanucleosides or any combination thereof. Such monomers may be coupled to each other by any of the numerous known internucleoside linkages. In certain preferred embodiments, these internucleoside linkages may be phosphodiester, phosphotriester, phosphorothioate, or phosphoramidate linkages, or combinations thereof. The linkages may be in any configuration, including without limitation 5′-3′, 5′-2′, 5′-5′, 3′-3′, 3′-5′, 2′-5′ or any combination thereof. The term "oligonucleotide" also encompasses such polymers having chemically modified bases or sugars and/or having additional substituents, including without limitation lipophilic groups, cholesterol, folic acid, intercalating agents, diamines and adamantane. Oligonucleotides amy also be formulated, *e.g.*, in cyclodextrins and/or liposomes. For purposes of the invention the term "2′-O-substituted" and 3′-O-substituted mean, respectively, substitution of the 2′ or 3′ position of the pentose moiety with a halogen (preferably Cl, Br, or F), or an -O-lower alkyl group containing 1-6 saturated or unsaturated carbon atoms, or with an -O-aryl or allyl group having 2-6 carbon atoms, wherein such alkyl, aryl or allyl group may be unsubstituted or may be substituted, e.g., with halo, hydroxy, trifluoromethyl, cyano, nitro, acyl, acyloxy, alkoxy, carboxyl, carbalkoxyl, or amino groups; or such 2′ substitution may be with a hydroxy group (to produce a ribonucleoside), an amino or a halo group, but not with a 2′-H group. In certain preferred embodiments, the 2′-O-substituted ribonucleoside is selected from 2′-O-methyl ribonucleosides and 2′-O-methoxyethoxy ribonucleosides. In certain preferred embodiments, the 3′-O-substituted ribonucleoside is selected from 3′-O-methyl ribonucleosides and 3′-O-methoxyethoxy ribonucleosides. In certain embodiments all nucleosides may be 2′-O-substituted, preferably 2′-O-alkyl. Oligonucleotides used in the methods according to the invention also include double stranded oligonucleotides, including hairpin oligonucleotides, as well as cyclic

oligonucleotides.

In certain preferred embodiments, an oligonucleotide for use in the invention may be complementary to an endogenous or exogenous nucleic acid sequence, preferably a nucleic acid that is involved in a disease. The term "complementary" means having the ability to hybridize to a genomic region, a gene, or an RNA transcript thereof under physiological conditions. Such hybridization is ordinarily the result of base-specific hydrogen bonding between complementary strands, preferably to form Watson-Crick or Hoogsteen base pairs, although other modes of hydrogen bonding, as well as base stacking can also lead to hybridization. As a practical matter, such hybridization can be inferred from the observation of specific gene expression inhibition. The nucleic acid sequence to which the modified oligonucleotide sequence is complementary will depend upon the biological effect that is sought to be modified. In certain particularly preferred embodiments the oligonucleotide is complementary to a gene selected from mdm-2, PKA, PKC, raf-kinase, bcl-2, H-ras, c-myc, DNA methyltransferase, histone deacetylase and VEGF. In certain preferred embodiments such an oligonucleotide has the sequence 5'-UGACACCTGTTCTCACUCAC-3'. However, in other preferred embodiments oligonucleotides having this sequence are specifically excluded, and in some preferred embodiments oligonucleotides that are complementary to the mdm-2 gene are specifically excluded. In certain embodiments, antisense oligonucleotides are specifically excluded, as the oligonucleotides used in the methods according to the invention are capable of potentiating the activity of prodrugs in a sequence independent manner.

Oligonucleotides in antisense embodiments are preferably from about 13 to about 100 nucleotides in length, more preferably from about 15 to about 50, and most preferably from about 15 to about 35. Oligonucleotides in non-antisense embodiments can be within these ranges, but can also preferably be from about 5 to about 15 nucleotides in length. Preferably, oligonucleotides used in the methods according to

the invention contain one or more modified internucleoside linkage and may optionally contain either deoxyribonucleosides, ribonucleosides or 2'-O-substituted ribonucleosides, or any combination thereof. Particularly preferred antisense oligonucleotides according to this aspect of the invention include mixed backbone oligonucleotides, including chimeric oligonucleotides and hybrid oligonucleotides.

For purposes of the invention, a "mixed backbone oligonucleotide" is an oligonucleotide having more than one type of backbone substituent, *e.g.*, differences in the sugar and/or internucleoside linkages among the various nucleosides comprising the oligonucleotide.

For purposes of the invention, a "chimeric oligonucleotide" refers to an oligonucleotide having more than one type of internucleoside linkage. One preferred embodiment of such a chimeric oligonucleotide is a chimeric oligonucleotide comprising a phosphorothioate, phosphodiester or phosphorodithioate region, preferably comprising from about 2 to about 12 nucleotides, and a nonionic region, preferably an alkylphosphonate or alkylphosphonothioate region. Preferably, such chimeric oligonucleotides contain at least three consecutive internucleoside linkages selected from phosphodiester and phosphorothioate linkages, or combinations thereof.

For purposes of the invention, a "hybrid oligonucleotide" refers to an oligonucleotide having more than one type of nucleoside. One preferred embodiment of such a hybrid oligonucleotide comprises a ribonucleotide or 2'-O-substituted ribonucleotide region, preferably comprising from about 2 to about 12 2'-O-substituted nucleotides, and a deoxyribonucleotide region. Preferably, such a hybrid oligonucleotide will contain at least three consecutive deoxyribonucleosides and will also contain ribonucleosides, 2'-O-substituted ribonucleosides, or combinations thereof. In a preferred embodiment, the deoxynucleotide region is flanked on either side by a 2'-O-substituted region. In one particularly preferred embodiment, the 2'-O-substituted regions are 2'-O-methyl regions, most preferably having four 2'-O-methyl

nucleosides. In certain preferred embodiments the entire backbone of the oligonucleotide is a phosphorothioate backbone. Particularly preferred hybrid oligonucleotides comprise one or more 2'-O-methyl ribonucleoside or 2'-O-methoxyethoxy ribonucleoside.

The synthesis of oligonucleotides can now be routinely accomplished. See e.g., *Methods in Molecular Biology, Vol 20: Protocols for Oligonucleotides and Analogs* pp. 165-189 (S. Agrawal, Ed., Humana Press, 1993); *Oligonucleotides and Analogues: A Practical Approach*, pp. 87-108 (F. Eckstein, Ed., 1991); and Uhlmann and Peyman, *supra*. Agrawal and Iyer, *Curr. Op. in Biotech.* 6: 12 (1995); and *Antisense Research and Applications* (Crooke and Lebleu, Eds., CRC Press, Boca Raton, 1993).

In a second aspect, the invention provides methods for statistically significantly potentiating the activity of a prodrug without producing significant side effects, the method comprising administering a polyanion prior to administration of the prodrug. It has been surprisingly discovered that administration of the polyanion prior to the administration of the prodrug results in even greater potentiation of the prodrug than when the polyanion is administered at the same time as, or after, administration of the prodrug.

Without wishing to be bound by theory, the potentiation of the prodrug is believed to involve one or more of the following mechanisms:

- modulation of the retention time of the prodrug in the liver and other tissues, including tumor tissue
- competition with cleavage enzymes or other hepatic enzymes, *e.g.*, carboxylesterases, amidases, or other esterases
- competition with transport factors from the liver, *e.g.*, cMOAT for CPT-11
- competition for binding of serum proteins

- competition with binding of endothelial cell walls
- competition for covalent modification, *e.g.*, glucouronidation
- slowing hydrolysis of the prodrug so that active metabolite is continuously released into the blood circulation
- stabilization of the active form of the drug, *e.g.*, lactone formation for CPT-11.

Any of these mechanisms may benefit by saturation of the system with the polyanion prior to administration of the prodrug.

The preferred embodiments of this aspect of the invention include all of the various embodiments discussed for the first aspect of the invention, and include the definitions set forth for the first aspect of the invention.

In a third aspect, the invention provides methods for statistically significantly potentiating the activity of a prodrug without producing significant side effects, the method comprising co-administering a polyanion with the prodrug, wherein the prodrug is present in an amount that would not be therapeutically effective in the absence of the polyanion. Methods according to this aspect of the invention are particularly useful where toxicity of the prodrug or active compound is dose-limiting. Thus the methods according to the invention can increase the therapeutic index for the prodrug.

The preferred embodiments of this aspect of the invention include all of the various embodiments discussed for the first and second aspects of the invention, and include the definitions set forth for the first and second aspects of the invention.

The following examples are intended to further illustrate certain particularly preferred embodiments of the invention and are not intended to limit the scope of the invention.

Example 1

Treatment of colon cancer tumor-bearing mice

Female NCr-nude mice, 6-8 weeks of age, were fed *ad libitum* water (reverse osmosis, 0.17% Cl) and an autoclaved standard rodent diet (NIH31) of 18% protein; 5% fat, 5% fiber, 8% ash and 3% minerals. Mice were housed in microisolators on a 12 hour light cycle at 22°C in 40-60% humidity. Mice were implanted subcutaneously in the flank with 1 mm^3 HCT-116 human colon carcinoma fragments in the flank. Tumors were monitored twice weekly initially, then daily as the tumors reached approximately 100 mg in weight. When the tumors reached a weight between 40-221 mg (calculated weight), the animals were pair-matched into the various treatment groups. Estimated tumor weight was determined according to the equation: tumor weight =

$$\frac{w^2 \times l}{2}$$

where w = width and l = length in mm of a HCT-116 tumor. Phosphorothioate oligonucleotides having 2′-O-methylribonucleosides at the 2 terminal 5′ positions and 4 terminal 3′ positions (Oligo 1), or the 4 terminal 5′ and 3′ positions (Oligo 2) were prepared according to standard procedures and dissolved in neutral buffered saline. Oligo 1 had the sequence 5′-UGACACCTGTTCTCACUCAC-3′ (complementary to mdm-2), and the aequence of Oligo 2 was 5′-UCGCACCCATCTCTCTCCUUC-3′ (complementary to the HIV-1 gag gene). Camptosar was purchased from Pharmacia & Upjohn.

Animals were pair-matched on Day 1 into 12 groups with 9 mice per group. Oligo or Oligo 2 was administered i.p. at 10mg/kg doses on a 5/2/5/2/5/2/5 schedule (*i.e.*, five days dosing, two days rest, repeat). Camptosar was administered i.v. at doses of 25 or 50 mg/kg once a week for 3 weeks. For combined treatments, 5 or 10 mg/kg of Oligo 1 was administered i.p. with Camptosar at 25 mg/kg, or 10 mg/kg Oligo 1 was administered was administered i.p. with 50 mg/kg Camptosar. Oligo 2

was administered at a dose of 10 mg/kg i.p. with 25 or 50 mg/kg Camptosar. Control animals were treated with vehicle i.p. on a 5/2/5/2/5/2/5 schedule. The study was terminated on day 56.

Results were determined using the tumor growth delay (TGD) endpoint method. Each mouse was euthanized when its HCT-116 tumor reached a weight of 1.5 g; this was taken as a cancer death. Mean Day of Survival (MDS) was calculated for each group based upon the calculated day of death according to: Time to end point (calculated) = Time to exceed endpoint (observed) minus

$$\frac{Wt_2 - \text{endpoint weight}}{\dfrac{Wt_2 - Wt_1}{D_2 - D_1}}$$

where Time to exceed endpoint (observed) is the number of days it takes for each tumor to grow past the endpoint (cut-off) weight (mouse is euthanized), D_2 is the day that the mouse is euthanized, D_1 is the last day of caliper measurement before the tumor reaches endpoint, Wt_2 is tumor weight (mg) on D_2, Wt_1 is tumor weight (mg) on D_1, and Endpoint weight is the predetermined "cut-off" tumor weight for the model being used. For statistical analysis, the unpaired t-test and Mann-Whitney U test (analyzing means and medians respectively) were used to determine the statistical significance of differences in survival times between groups. These analyses were conducted at a p level of 0.05 (two-tailed) using Prism (GraphPad) version 3.0.

Of the 9 vehicle control mice, 8 had tumors reaching the 1.5 g endpoint with an MDS value of 21.5 days. One tumor regressed completely, presumably due to poor tumor take. Camptosar at 25 mg/kg produced an MDS value of 31.1 days, and at 50 mg/kg, 42.6 days. Neither Oligo 1 nor Oligo 2 alone produced any prolongation of MDS. However, administration of 10 mg/kg Oligo 1 with 25 mg/kg Camptosar extended MDS over vehicle controls by 24.4 days, and over mice treated with 25 mg/kg Camptosar alone by 14.8 days. Each of these extensions is statistically

significant ($p < 0.0001$; unpaired t-test). Mice treated with 5 mg/kg Oligo 1 and 25 mg/kg achieved an MDS value of 37.4 days, which was statistically significant over vehicle controls ($p < 0.0005$; unpaired t-test) and over mice treated with 25 mg/kg Camptosar alone ($p < 0.046$; unpaired t-test). Administration of 10 mg/kg Oligo 2 i.p. with 25 mg/kg Camptosar produced an MDS value of 39.7 days, which is statistically significant over vehicle controls ($p < 0.0001$; unpaired t-test) and over mice treated with 25 mg/kg Camptosar alone ($p < 0.0009$; unpaired t-test). Administration of 10 mg/kg Oligo 2 i.p. with 50 mg/kg Camptosar produced an MDS value of 42.6 days, which trends toward statistical significance over mice treated with 50 mg/kg Camptosar alone ($p < 0.08$; unpaired t-test). These results demonstrate that both Oligo 1 and Oligo 2 can potentiate the activity of Camptosar efficacy in a statistically significant and dose-dependent manner, and that at least part of this effect is independent of oligonucleotide sequence. The results of these studies are summarized in Figures 1-4.

Comparison of the potentiation of Camptosar efficacy by Oligo 1 against potentiation of Camptosar efficacy by Oligo 2 shows that there is a statistically significant difference in favor of Oligo 1 ($p < 0.0074$; unpaired t-test). It is believed that this difference may arise from an antisense effect of Oligo 1 on expression of the mdm-1 oncogene to which it is complementary.

Example 2

Treatment of pancreatic cancer tumor-bearing mice

To test whether the differences between Oligo 1 and Oligo 2 resulted from an antisense effect by Oligo 1, similar studies were conducted in a mouse model for pancreatic cancer (Panc 1 tumor). The Panc 1 tumor has a mutant (nonfunctional) p53 gene. Since antisense effects against mdm-1 are believed to work primarily by upregulating p53 expression, Oligo 1 should not produce an antisense specific effect in

this model. However, it is possible that mdm-2 targeted oligonucleotides in p53 mutant cell lines may work by a mechanism independent of p53.

The study was carried out as described in Example 1, except that Panc-1 tumor was used, 4 groups of 10 mice each were used, Oligo 1 and Oligo 2 (in this case, 5'-UCCCACCTATTCTTACUCCC-3', with two 5'-terminal 2'-O-methylribonucleosides and four 3'-terminal 2'-O-methylribonucleosides) were given at doses of 20 mg/kg, Camptosar was given at 100 mg/kg, tumor "cut-off" was 1.2 g, and the study was terminated on Day 67.

Both Oligo 1 and Oligo 2 showed statistically significant potentiation of Camptosar efficacy ($p < 0.05$; unpaired t-test). The potentiating effects of Oligo 1 and Oligo 2, compared with each other, were statistically indistinguishable. These results demonstrate that oligonucleotides produce a statistically significant potentiating effect on Camptosar that is independent of the sequence of the oligonucleotide. Moreover, in these studies treatment with Camptosar alone was not statistically significantly better than treatment with vehicle. Thus, these results demonstrate that oligonucleotides can potentiate the effectiveness of Camptosar such that an otherwise sub-therapeutic dosage of Camptosar becomes therapeutically effective.

Example 3

Effect of timing and route of oligonucleotide administration

The study of Example 1 was repeated, but the oligonucleotide was administered initially on day 1 and Captosar was not administered initially until day 3. Surprisingly, this schedule of administration was even more effective (see Figure 5). Also, the study of Example 1 was repeated, but the oligonucleotide was administered orally. This route of administration was equally effective (see Figure 6).

What is claimed is:

1. A method for statistically significantly potentiating the activity of a prodrug without producing significant side effects, the method comprising co-administering a polyanion with the prodrug, wherein the polyanion is not an oligonucleotide having two 5′ and four 3′ 2′-O-methylribonucleosides and having the sequence 5′-UGACACCTGTTCTCACUCAC-3′.

2. The method according to claim 1, wherein the prodrug is an ester or an amide of an active compound.

3. The method according to claim 2, wherein the active compound is an anti-cancer drug.

4. The method according to claim 3, wherein the anti-cancer drug is SN-38.

5. The method according to claim 4, wherein the prodrug is Camptosar.

6. The method according to any of claims 1-5, wherein the polyanion is a selected from polysulfates and oligonucleotides.

7. The method according to claim 6, wherein the polysulfate is selected from heparin, dextran sulfates, suramin sulfates, cyclodextrin sulfates and oligonucleotide phosphorothioates or phosphorodithioates.

8. The method according to claim 7, wherein the oligonucleotide comprises a 2′-O-substituted ribonucleoside.

9. The method according to claim 8, wherein the 2'-O-substituted ribonucleoside is selected from 2'-O-methyl ribonucleosides and 2'-O-methoxyethoxy ribonucleosides.

10. A method for statistically significantly potentiating the activity of a prodrug without producing significant side effects, the method comprising co-administering a polyanion with the prodrug,, wherein the polyanion is administered before the prodrug.

11. The method according to claim 10, wherein the prodrug is an ester or an amide of an active compound.

12. The method according to claim 11, wherein the active compound is an anti-cancer drug.

13. The method according to claim 12, wherein the anti-cancer drug is SN-38.

14. The method according to claim 13, wherein the prodrug is Camptosar.

15. The method according to any of claims 10-14, wherein the polyanion is a selected from polysulfates and oligonucleotides.

16. The method according to claim 15, wherein the polysulfate is selected from heparin, dextran sulfates, suramin sulfates, cyclodextrin sulfates and oligonucleotide phosphorothioates or phosphorodithioates.

17. The method according to claim 15, wherein the oligonucleotide

comprises a 2'-O-substituted ribonucleoside.

18. The method according to claim 17, wherein the 2'-O-substituted ribonucleoside is selected from 2'-O-methyl ribonucleosides and 2'-O-methoxyethoxy ribonucleosides.

19. A method for statistically significantly potentiating the activity of a prodrug without producing significant side effects, the method comprising co-administering a polyanion with the prodrug,, wherein the prodrug is present in an amount that would not be therapeutically effective in the absence of the polyanion.

20. The method according to claim 19, wherein the prodrug is an ester or an amide of an active compound.

21. The method according to claim 20, wherein the active compound is an anti-cancer drug.

22. The method according to claim 21, wherein the anti-cancer drug is SN-38.

23. The method according to claim 22, wherein the prodrug is Camptosar.

24. The method according to any of claims 19-23, wherein the polyanion is a selected from polysulfates and oligonucleotides.

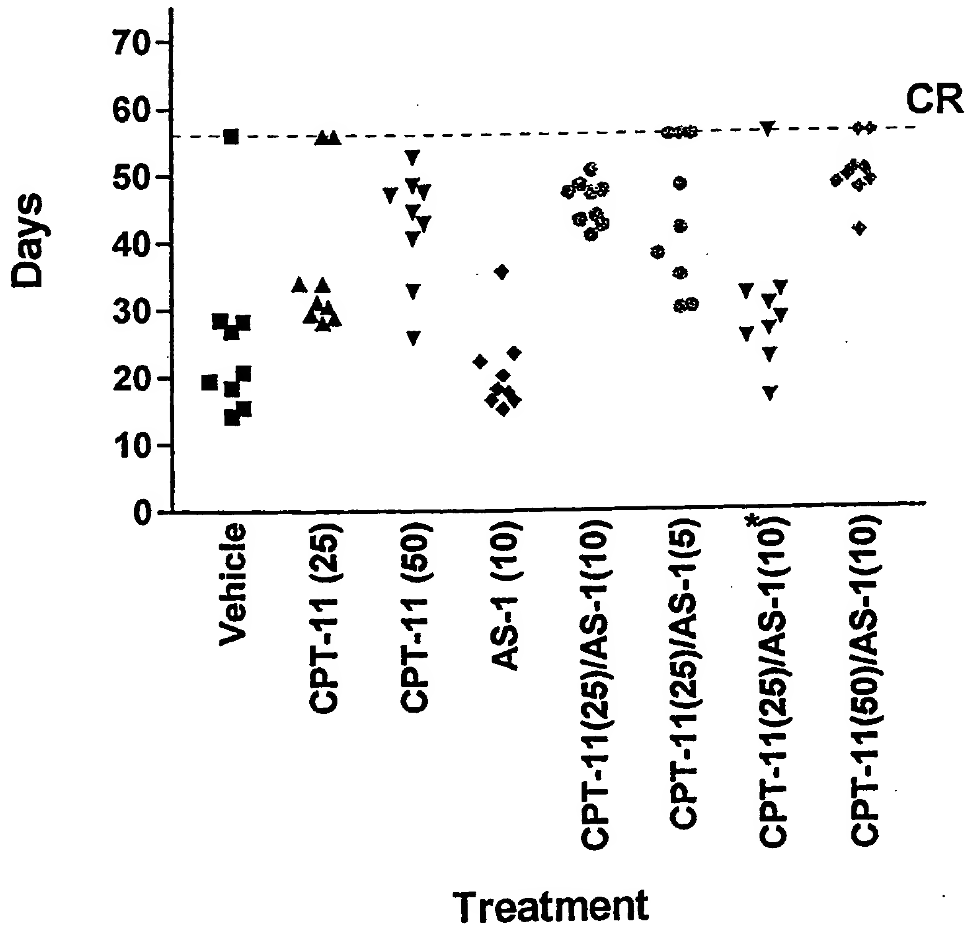
25. The method according to claim24, wherein the polysulfate is selected from heparin, dextran sulfates, suramin sulfates, cyclodextrin sulfates and oligonucleotide phosphorothioates or phosphorodithioates.

26. The method according to claim 25, wherein the oligonucleotide comprises a 2'-O-substituted ribonucleoside.

27. The method according to claim 26, wherein the 2'-O-substituted ribonucleoside is selected from 2'-O-methyl ribonucleosides and 2'-O-methoxyethoxy ribonucleosides.

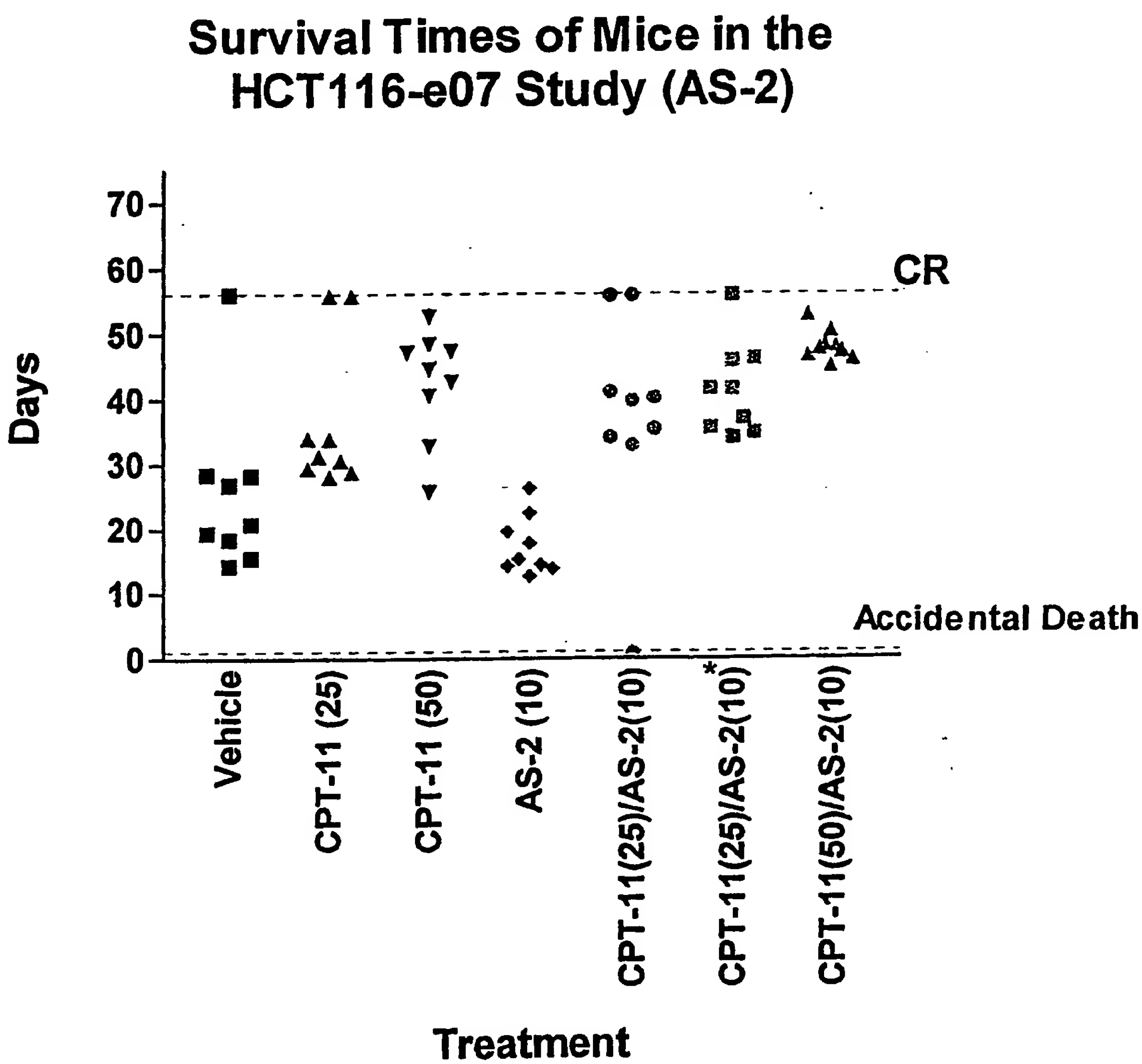
Figure 1

Survival Times of Mice in the HCT116-e07 Study (AS-1)



*p.o.

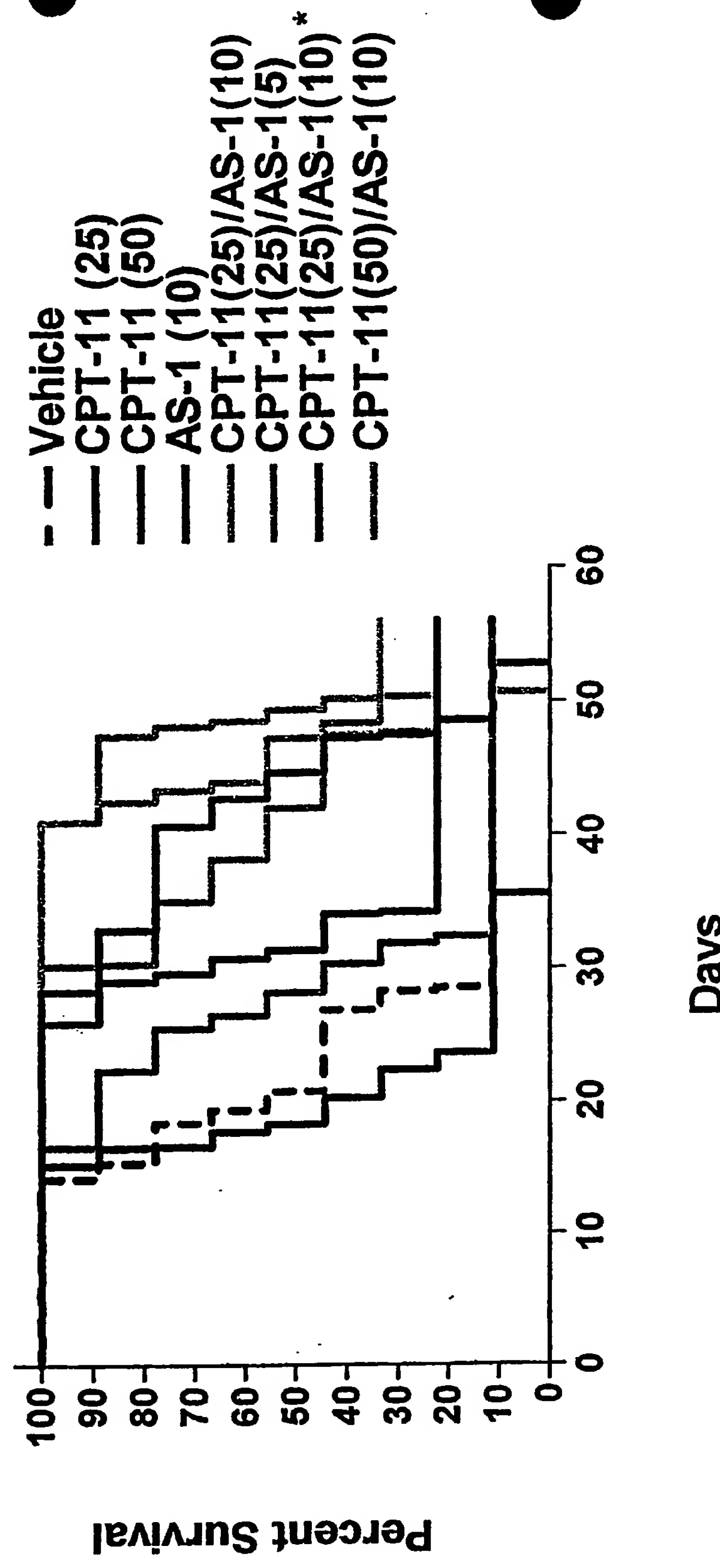
Figure 2


Survival Times of Mice in the HCT116-e07 Study (AS-2)
Days
70
60
50
40
30
20
10
0
CR
Accidental Death
Vehicle
CPT-11 (25)
CPT-11 (50)
AS-2 (10)
CPT-11(25)/AS-2(10)
*CPT-11(25)/AS-2(10)
CPT-11(50)/AS-2(10)
Treatment

*p.o.

Figure 3

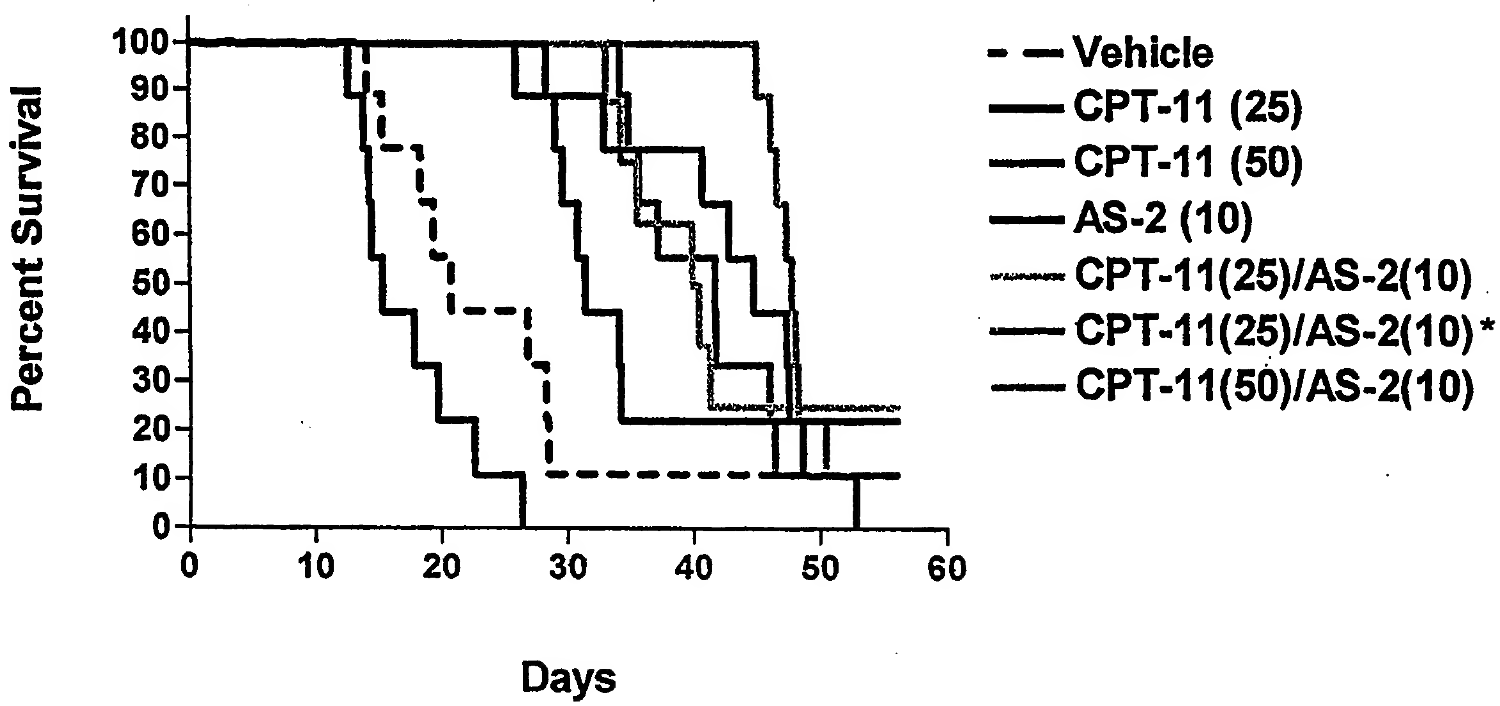
Kaplan-Meier Survival Plot for Mice in the HCT116-e07 Study (AS-1)



*p.o.

Figure 4

Kaplan-Meier Survival Plot for Mice in the HCT116-e07 Study (AS-2)



*p.o.

Figure 5

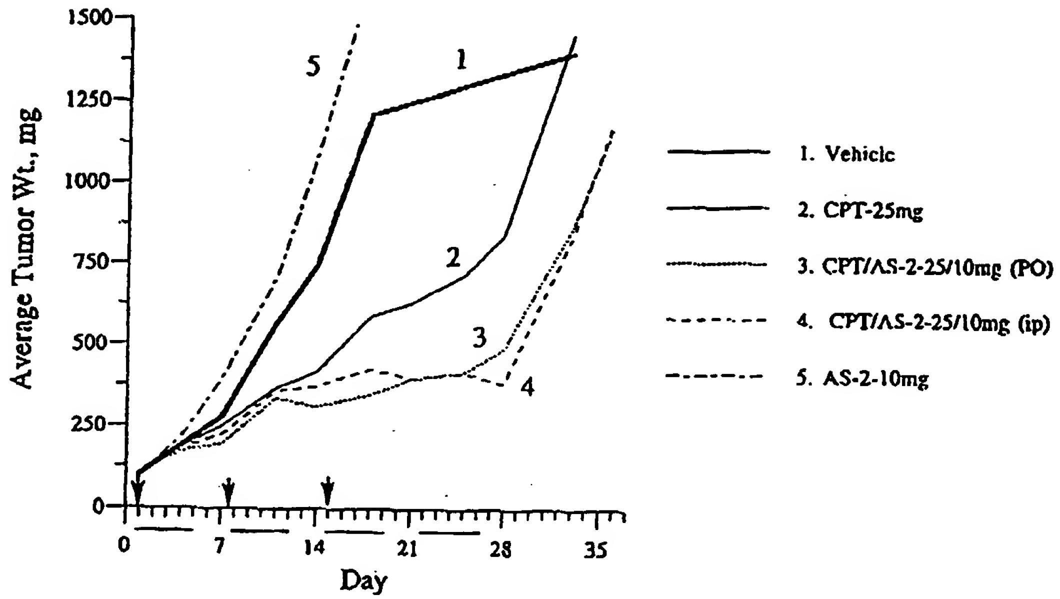
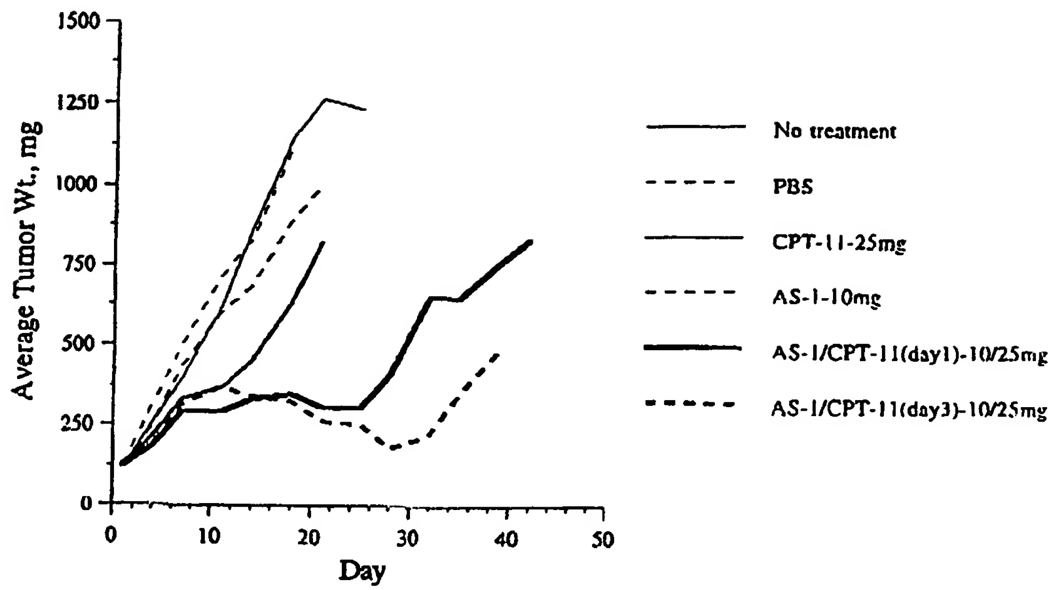

1500
1250
1000
750
500
250
0
Average Tumor Wt., mg
0
7
14
21
28
35
Day
5
1
2
3
4
1. Vehicle
2. CPT-25mg
3. CPT/AS-2-25/10mg (PO)
4. CPT/AS-2-25/10mg (ip)
5. AS-2-10mg

Figure 6


1500
1250
1000
750
500
250
0
Average Tumor Wt., mg
0
10
20
30
40
50
Day
No treatment
PBS
CPT-11-25mg
AS-1-10mg
AS-1/CPT-11(day1)-10/25mg
AS-1/CPT-11(day3)-10/25mg